PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant(s): Judith E. Kimble
Robert H. Blelloch

Date: _____

Serial No.: 09/321,987

Group Art Unit: 1632

Filed: May 28, 1999

Examiner: R. Shukla

For: AGENT AND METHOD FOR MODULATION OF CELL MIGRATION

File No.: 960296.95386

DECLARATION UNDER 37 C.F.R. §1.132

Commissioner For Patents
Washington DC 20231

Dear Sir:

I, Judith E. Kimble, on oath say and declare that:

1. I am the same Judith E. Kimble who is one of the named inventors of the above-identified patent application. I am currently employed by the Howard Hughes Medical Institute at the University of Wisconsin-Madison where I am a Professor of Biochemistry, Molecular Biology, and Medical Genetics. I have worked as a research scientist specializing in the general area of developmental biology. I have studied development in nematodes for 28 years. I have published extensively in the area and have published several state-of-the-art review articles on this subject. I was inducted into the National Academy of Sciences of the United States of America in 1995.

2. I have reviewed the Office Actions issued in this matter by the U.S. Patent and Trademark Office on December 13, 2000 and on July 3, 2001. I understand that Claims 1-13 are rejected for alleged lack of enablement, in part because the Examiner was not convinced that proteins other than native GON-1 can actively direct migration of a gonadal distal tip cell in a nematode. This Declaration is submitted to provide evidence that other proteins can have such an activity in a nematode.

3. At my direction, and under my supervision, Dr. Craig Newman, a post-doctoral fellow in my laboratory, demonstrated in the manner described below that a human aggrecan-

degrading metalloprotease ("aggrecanase") has a distal tip cell migration-directing activity in a nematode, as was predicted in the patent application.

4. A polynucleotide that encodes an aggrecanase protein in humans was placed under the control of the lag-2 transcriptional promoter in a plasmid vector thereafter designated pJK753. The lag-2 promoter is operable and promotes expression in nematode distal tip cells, as was described by Henderson et al., *Development,* 120:2913-2924 (1994). The polynucleotide that encodes the human aggrecanase protein was described by Tortorella, M.D., et al., "Purification and Cloning of Aggrecanase-1: A Member of the ADAMTS Family of Proteins," Science 284:1664 (1999). The sequence of the polynucleotide was disclosed in International Patent Application No. PCT/US98/15438 (Publication No. WO 99/05291), incorporated by reference into the subject US patent application (serial number 09/321,987) at page 15, line 9. The sequence is also available at GenBank Accession Number AF148213. The plasmid vector is otherwise conventional.

5. Transgenic *C. elegans* individuals were created by introducing plasmid pJK753 into *unc-24 gon-1/dpy-20* heterozygotes using the well-known DNA microinjection method for generating transgenic nematodes. From these injections, multiple transgenic lines were obtained.

6. Gonadal arm extension resulting from distal tip cell migration was evaluated in *gon-1* mutants and in *gon-1* transgenic mutants that produce human aggrecanase and have extrachromosomal arrays carrying the aggrecanase-encoding polynucleotide sequence. The arm extension migration assay is as described in Blelloch, R. and J. Kimble, "Control of organ shape by a secreted metalloprotease in the nematode *Caenorhabditis elegans*," Nature 399:586-590 (1999), already made of record in connection with the subject patent application.

7. Among the transgenic *unc-24 gon-1* mutants, partial gonadal arm extension was observed in multiple animals: arm extension was observed in 30% (n=10) and 17% (n=6) of the transgenic animals. Although partially rescued for arm extension, none of the transgenic *gon-1* mutants was fertile, demonstrating that the transgenic animals were not recombinants.

8. In contrast, no gonadal arm extension was observed in any parent animal.

9. I hereby declare that all statements made herein of my own knowledge are true, and that all statements made on information and belief are believed to be true; and further, that these statements are made with knowledge that willful false statements, and the like so made, are punishable by fine or imprisonment, or both, under Section 1001, Title 18 of the

United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Dated this 30th day of December 01.

Judith E. Kimble